

Chapter 1

General Introduction

The phylogenetic position of the Bryozoa is subject to ongoing discussions. Bryozoa or Ectoprocta are a group of sessile, colonial invertebrates which occur, with probably 8,000 recent species worldwide, in marine and limnetic habitats (introductory reviews in Ryland 1970, 2005, Mukai et al. 1997, Nielsen 2001). Since Hatschek (1891), the predominant opinion was that they form, together with Brachiopoda and Phoronida, the Tentaculata or Lophophorata (Hyman 1959). These were assigned either to protostomes or to deuterostomes or were placed “intermediate”. Despite varying views about the monophyly of lophophorates, most later approaches united them with Deuterostomia as Radialia (Jefferies 1986, Eernisse et al. 1992, Backeljau et al. 1993, Ax 1989, Brusca and Brusca 2003). This was based chiefly on the coelomic organization and the apparent homology of the tentacular apparatus to that of hemichordates and crinoid echinoderms. An alternative hypothesis, based mainly on larval morphology and similarities in metamorphosis, has been put forward by (Nielsen 1971, 1977, 2001, 2002), who agreed to the deuterostome affinity of Brachiopoda and Phoronida, but suggested a sister-group relationship between Bryozoa and Kamptozoa, thus a protostome affinity of bryozoans.

In a phylogenetic study using 18S rDNA data, Halanych et al. (1995) revealed evidence for a common ancestry of the lophophorate taxa, annelids and molluscs united in a clade named Lophotrochozoa. This new evidence challenged the long-held view of a deuterostome relationship of all or some lophophorate groups and many of the ideas that supported this. Subsequent studies of more extensive datasets and further molecular markers almost consistently corroborated Lophotrochozoa (see Halanych 2004 for review). Since at the same time many cladistic analyses of broad morphological datasets still supported deuterostome affinities (Zrzavy et al. 1998, 2003, Sorensen et al. 2000, Peterson and Eernisse 2001, Nielsen 2001, Glenner et al. 2004), a serious conflict appeared. However, until presently molecular approaches either fail to resolve the exact position of Bryozoa (as well as that of several other lophotrochozoan taxa) or provide conflicting evidences. Bryozoa have, for example, been suggested to be basal Lophotrochozoans (Halanych et al. 1995, Passamanek and Halanych 2006), basal protostomes (Giribet et al. 2000), polyphyletic (Mackey et al. 1996, Passamanek and Halanych 2006), polyphyletic with phoronid affinities (Helmkampf et al. 2008), sister to Brachiopoda (Waeschenbach et al. 2006), sister to Kamptozoa (Hausdorf et al. 2007).

Concerning the molecular data, several sources of error have been suggested that could cause the current problems (Telford et al. 2005, e.g., Philippe and Telford 2006, Baurain et al. 2007). Although some authors (Scotland et al. 2003) question in principle the value of morphology for phylogenetic inference, most researchers agree that morphology represents a complex data set whose careful analysis can serve as an independent test for molecular based phylogenies (Wiens 2004, Jenner 2004). Morphology-based phylogenies depend heavily on the characters chosen and the accuracy of the used details (Jenner and Schram 1999). Morphological data sets suffer chiefly from two problems. One is that the number of phylogenetically informative characters is relatively low in comparison to DNA sequence data. Especially in “phylum-level” phylogenies of the metazoa only few characters are comparable across the different groups. Thus, a single character has potentially a relatively high impact on the result of the analysis. This poses the second problem: Many characters are only insufficiently investigated in certain groups, leading to a high degree of uncertainty, when coding character states in a data matrix. This also applies for Bryozoa: looking at the most recent morphological data matrices that have been used for analyzing metazoan phylogeny (Zrzavy et al. 1998, 2003, Sorensen et al. 2000, Peterson and Eernisse 2001, Nielsen 2001, Glenner et al. 2004), it turns out that a majority of those characters that can be suspected to contain essential information, are scored “unknown” for Bryozoa. This is especially the case for characters of embryogenesis, neural organization and mesodermal structures. In contrast to this situation, detailed comparable data from many other metazoa taxa have become available by a growing field of comparative developmental biology and neuroanatomy during the last years (see e.g., Nielsen 2004, 2005, Minelli and Fusco 2008, Lichtneckert and Reichert 2007). Hence, in this thesis, a selection of these characters have been investigated with the central aim to provide new phylogenetically significant data.

Commonly the three bryozoan subtaxa Phylactolaemata, Stenolaemata, and Gymnolaemata, which differ in terms of both morphology and life-style, are distinguished. Gymnolaemata comprise Cheilostomata and Ctenostomata. In the traditional phylogenetic hypotheses, the morphology of Phylactolaemata has been regarded as especially significant, because several similarities to Phoronida had been recognized. Indeed, Phylactolaemata differ considerably from Stenolaemata and Gymnolaemata, hindering assumptions about ancestral character states for Bryozoa. Chapter 2 *Ganglion ultrastructure in phylactolaemate Bryozoa: Evidence for a neuroepithelium*, addresses the question whether the cerebral ganglion in Phylactolaemata is, in contrast to that of Gymnolaemata, hollow and formed by an invagination process comparable to the vertebrate neurulation. Chapter 3: *Ultrastructure of the body cavities in phylactolaemate Bryozoa* aims to answer the question, whether the epistome, an upper-lip like organ contains an independent secondary body cavity, as implied by earlier morphologists.

The evolution of larval types within Bryozoa is still elusive. Stenolaemata and Phylactolaemata exhibit larvae, which are hardly comparable to those of gymnolaemates as well as other metazoan groups (Reed 1991). In gymnolaemates, the planktotrophic cyphonautes, as well as several types of lecithotrophic larvae occur (Zimmer and Woollacott 1977). Chapter 4 *Muscular systems in gymnolaemate bryozoan larvae* focuses on the phylogenetic significance of larval musculature for the evolution of bryozoan larval types. During the last ten years, larval neural anatomy has regained much attention as complex character, of possibly high phylogenetic significance. Only few results on bryozoan larvae exist, which do not provide a consistent picture by now. Thus, in chapter 5 *Serotonergic and FMRFamidergic nervous systems in gymnolaemate bryozoan larvae*, data on nervous system characters of different larvae are collected and a putative ancestral pattern is proposed.

Early embryology in Bryozoa has been studied by several earlier workers, but the origin of mesoderm is still essentially unknown. As data on mesoderm formation have been acquired in many metazoan taxa, namely candidate sister groups like Phoronida and Brachiopoda (Freeman 2000, 2003, Lüter 2000, Freeman and Martindale 2002), knowledge on this character in Bryozoa has the potential to contribute arguments for the phylogenetic position of Bryozoa. In chapter 6 *Mesoderm origin in Membranipora membranacea*, developmental stages of a species exhibiting a cyphonautes larva are examined on the ultrastructural level.

Chapter 7 summarizes the most important results of this thesis and discusses their impact on the conceptions about phylogeny and position of the Bryozoa .

References

- Ax P (1989) Basic phylogenetic systematization of the Metazoa. In: Fernholm B, Bremer K, Jornvall H (eds) *The Hierarchy of Life*. Elsevier, New York, pp 229-245
- Backeljau T, Winnepenninckx B, Debruyne L (1993) Cladistic Analysis of Metazoan Relationships - A Reappraisal. *Cladistics* 9:167-181
- Baurain D, Brinkmann H, Philippe H (2007) Lack of resolution in the animal phylogeny: Closely spaced cladogeneses or undetected systematic errors? *Molecular Biology and Evolution* 24:6-9
- Brusca RC, Brusca GJ (2003) *Invertebrates*. Sinauer Associates, Sunderland, Mass
- Eernisse DJ, Albert JS, Anderson FE (1992) Annelida and Arthropoda Are Not Sister Taxa - A Phylogenetic Analysis of Spiralian Metazoan Morphology. *Syst Biol* 41:305-330
- Freeman G (2000) Regional specification during embryogenesis in the inarticulate brachiopod *Crania anomala*. *Dev Biol* 227:219-238
- Freeman G (2003) Regional specification during embryogenesis in Rhynchonelliform brachiopods. *Dev Biol* 261:268-287

- Freeman G, Martindale MQ (2002) The origin of mesoderm in phoronids. *Dev Biol* 252:301-311
- Giribet G, Distel DL, Polz M, Sterrer W, Wheeler WC (2000) Triploblastic Relationships with Emphasis on the Acoelomates and the Position of Gnathosomulida, Ciliophora, Plathelminthes and Chaetognatha: A Combined Approach of 18S rDNA Sequences and Morphology. *Syst Biol* 49:539-562
- Glenner H, Hansen AJ, Sorensen MV, Ronquist F, Huelsenbeck JP, Willerslev E (2004) Bayesian Inference of the Metazoan Phylogeny: A Combine Molecular and Morphological Approach. *Curr Biol* 14:1644-1649
- Halanych KM (2004) The new view of animal phylogeny. *Ann Rev Ecol Syst* 35:229-256
- Halanych KM, Bacheller JD, Aguinaldo AMA, Liva SM, Hills DM, Lake JA (1995) Evidence from 18S ribosomal DNA that the lophophorates are protostome animals. *Science* 267:1641-1643
- Hatschek B (1891) *Lehrbuch der Zoologie*. Gustav Fischer, Jena
- Hausdorf B, Helmkampf M, Meyer A, Witek A, Herlyn H, Bruchhaus I, Hankeln T, Struck TH, Lieb B (2007) Spiralian phylogenomics supports the resurrection of Bryozoa comprising Ectoprocta and Entoprocta. *Molecular Biology and Evolution* 24:2723-2729
- Helmkampf M, Bruchhaus I, Hausdorf B (2008) Multigene analysis of lophophorate and chaetognath phylogenetic relationships. *Mol Phylogenet Evol* 46:206-214
- Hyman LH (1959) *The Invertebrates V: Smaller Coelomate Groups*. McGraw-Hill, New York
- Jefferies RPS (1986) *The ancestry of vertebrates*. British Museum (Natural History), London
- Jenner RA (2004) Accepting partnership by submission? Morphological phylogenetics in a molecular millennium. *Syst Biol* 53:333-342
- Jenner RA, Schram FR (1999) The grand game of metazoan phylogeny: rules and strategies. *Biol Rev* 74:121-142
- Lichtneckert R, Reichert H (2007) Origin and evolution of the first nervous system. In: Kaas JH (eds) *Evolution of Nervous Systems Vol. 1: Theories, Development, Invertebrates*. Academic Press, 289-315
- Lüter C (2000) The origin of the coelom in Brachiopoda and its phylogenetic significance. *Zoomorphology* 120:15-28
- Mackey LY, Winnepeninckx B, De Wachter R, Backeljau T, Emschermann P, Garey JR (1996) 18s rRNA Suggests That Entoprocta Are Protostomes, Unrelated to Ectoprocta. *J Mol Evol* 42:552-559
- Minelli A, Fusco G (2008) *Evolving Pathways: Key Themes in Evolutionary Developmental Biology*. Cambridge University Press, Cambridge
- Mukai H, Terakado K, Reed CG (1997) Bryozoa. In: Harrison FW, Woollacott RM (eds) *Microscopic Anatomy of Invertebrates Vol. 13: Lophophorates, Entoprocta and Ciliophora*. Wiley-Liss, New York, pp 45-206
- Nielsen C (2002) The phylogenetic position of Entoprocta, Ectoprocta, Phoronida, and Brachiopoda. *Integ Comp Biol* 42:685-691

- Nielsen C (2005) Trochophora larvae: Cell-lineages, ciliary bands, and body regions. 2. Other groups and general discussion. *J Exp Zool B* 304:401-447
- Nielsen C (1977) The relationships of Entoprocta, Ectoprocta and Phoronida. *Amer Zool* 17:149-150
- Nielsen C (2001) Animal evolution. Interrelationships of the living phyla. Oxford University Press,
- Nielsen C (1971) Entoproct life-cycles and the entoproct/ectoproct relationship. *Ophelia* 9:209-341
- Nielsen C (2004) Trochophora larvae: Cell-lineages, ciliary bands, and body regions. 1. Annelida and Mollusca. *J Exp Zool B* 302:35-68
- Passamanek YJ, Halanych KM (2006) Lophotrochozoan phylogeny assessed with LSU and SSU data: Evidence of lophophorate polyphyly. *Mol Phylogenet Evol* 40:20-28
- Peterson KJ, Eernisse DJ (2001) Animal phylogeny and the ancestry of bilaterians: inferences from morphology and 18S rDNA gene sequences. *Evol Dev* 3:170-205
- Philippe H, Telford MJ (2006) Large-scale sequencing and the new animal phylogeny. *Trends in Ecology & Evolution* 21:614-620
- Reed CG (1991) Bryozoa. In: Giese AC, Pearse JS, Pearse VB (eds) *Reproduction of Marine Invertebrates VI Echinoderms and Lophophorates*. Boxwood Press, Pacific Grove, California, pp 85-245
- Ryland JS (1970) *Bryozoans*. Hutchinson University Library, London
- Ryland JS (2005) Bryozoa: an introductory overview. *Denisia* 16:9-20
- Scotland RW, Olmstead RG, Benett JR (2003) Phylogeny reconstruction: the role of morphology. *Syst Biol* 52:539-548
- Sorensen MV, Funch P, Willerslev E, Hansen AJ, Olesen J (2000) On the Phylogeny of the Metazoa in the Light of Cycliophora and Micrognathozoa. *Zool Anz* 239:297-318
- Telford MJ, Wise MJ, Gowri-Shankar V (2005) Consideration of RNA secondary structure significantly improves likelihood-based estimates of phylogeny: Examples from the bilateria. *Molecular Biology and Evolution* 22:1129-1136
- Waeschenbach A, Telford MJ, Porter JS, Littlewood DTJ (2006) The complete mitochondrial genome of *Flustrellidra hispida* and the phylogenetic position of Bryozoa among the Metazoa. *Mol Phylogenet Evol* 40:195-207
- Wiens JJ (2004) The role of morphological data in phylogeny reconstruction. *Syst Biol* 53:653-661
- Zimmer RL, Woollacott RM (1977) Structure and classification of gymnolaemate larvae. In: Woollacott RM, Zimmer RL (eds) *Biology of bryozoans*. Academic Press, New York, pp 57-89
- Zrzavy J (2003) Gastrotricha and metazoan phylogeny. *Zool Scr* 32:61-81
- Zrzavy J, Mihulka S, Kepka P, Bezdek A, Tietz D (1998) Phylogeny of the Metazoa based on morphological and 18S ribosomal DNA evidence. *Cladistics* 14:249-285